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Evaluation for Multiple Pest Resistance in European Corn Borer, *Ostrinia nubilalis*, Resistant Maize Accessions from Peru

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ABSTRACT: Eleven maize, *Zea mays* L., accessions from Peru previously found resistant to leaf feeding by first generation European corn borer (ECB1), *Ostrinia nubilalis* (Hübner), were evaluated for resistance to stalk boring by second generation European corn borer (ECB2), southwestern corn borer (SWCB), *Diatraea grandiosella* Dyar, fall armyworm (FAW), *Spodoptera frugiperda* (J. E. Smith), western corn rootworm (WCRW), *Diabrotica virgifera virgifera* LeConte, corn earworm (CEW), *Helicoverpa zea* (Boddie), and sugarcane borer (SCB), *Diatraea saccharalis* (Fab.). Four accessions were resistant to 2 pests in addition to ECB1: PI 503720 was resistant to WCRW and ECB2, whereas PI 503728, PI 503849, and A-10623 were resistant to CEW and ECB2. Accession PI 503720 also had intermediate resistance to CEW and SCB. Accessions PI 503728 and PI 503849 had intermediate resistance to WCRW and SCB. Accession A-10623 had intermediate resistance to CEW larval weight and days to pupation and to SCB. Multiple pest resistance in maize, such as that detected in this study, would be useful in pest management, especially in sustainable agriculture systems.

Abel and Wilson (1995) evaluated all available accessions of Peruvian maize, *Zea mays* L., maintained within the U.S. National Plant Germplasm System for leaf feeding (first generation) resistance to European corn borer (ECB), *Ostrinia nubilalis* (Hübner), and found 11 resistant accessions. All 11 accessions are early, drought resistant, floury-type maize originating at lower elevations along Peru's north coast. Current germplasm sources of ECB leaf-feeding resistance are based primarily on the chemical DIMBOA (2,4-dihydroxy-7-methoxy-1,4-benzoxazin-3-one) in the whorl-leaf tissue. The ECB resistance in the 11 Peruvian maize accessions is not based on DIMBOA but caused by other unknown factor(s) (Abel and Wilson, 1995).

The purpose of this study was to determine if the 11 Peruvian maize accessions also possess resistance to other maize insect pests, such as with the multiple insect resistance observed by Davis et al. (1988) in maize from Antigua. The 11 acces-

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sions included in this study were evaluated for resistance to second generation ECB, southwestern corn borer (SWCB), *Diatraea grandiosella* Dyar, fall armyworm (FAW), *Spodoptera frugiperda* (J. E. Smith), corn earworm (CEW), *Helicoverpa zea* (Boddie), western corn rootworm (WCRW), *Diabrotica virgifera virgifera* LeConte, and sugarcane borer (SCB), *Diatraea saccharalis* (Fabricius).

Materials and Methods

The 11 Peruvian maize accessions were planted in Ames, IA; Tifton, GA; State College, MS; Columbia, MO; and Houma, LA, during the summers of 1993 and 1994. Standard maize production procedures were practiced for the areas in which they were planted.

At Ames, the accessions were evaluated for second-generation ECB. The 11 accessions and a resistant check inbred, B-52, were grown in a randomized block design replicated 4 times. Rows were 7.6 m long and 0.9 m apart. Ten plants per replication were infested at anthesis with 300 neonate ECB larvae. Three weeks after infestation, the plants were cut at ground level, split lengthwise, and the length of tunnelling caused by ECB was measured. Data were analyzed with the ANOVA-2 program of MSTAT-C (MSTAT Development Team, 1989). When the F-value for treatments was significant ($P < 0.05$), means were separated with the least significant difference (LSD) test ($\alpha = 0.05$) included in the RANGE program of MSTAT-C.

At Tifton, the plants were evaluated for resistance to silk-feeding by CEW. The 11 accessions, plus a resistant check 'Zapalote Chico 2451# (P) (C3)' (ZC), and a susceptible check 'Stowell's Evergreen' (SEG), were planted in two row plots 6.1 m long and 0.9 m apart. Ear shoots were covered on all plants. After silks had emerged for 2 d, nonpollinated silks were excised to the ear tip, taken to the laboratory, and removed from the silk channel. Silks were bulked from each entry, oven-dried for 10 days at 41°C, and ground (1-mm screen) with a Cyclotec sample mill (Fisher Scientific, Atlanta, GA). Then, 20 g of dry silk were mixed in 400 ml dilute (3 ml diet : 2 ml water) pinto bean diet for each entry. A pinto bean diet check was included that also had 20 g of celufil. The silk-diet mixtures for each entry were dispensed (10 ml/cup) into 30-ml plastic cups and allowed to cool for 2 h. Subsequently, one neonate CEW was placed in each cup, and the cup capped. The experiment was arranged as a randomized complete-block design with 30 replications (1 cup/replicate). The experiment was held in a controlled-environment room at 26.7 ± 2°C, 70 ± 5% RH with a photoperiod of 14:10 (LD). Weight (mg) of larvae was recorded 8 d after infestation. Also, development time (d) and weight (mg) of pupae were recorded. Data were analyzed by using the GLM Procedure of SAS (SAS Institute, 1989), and means were separated by Waller-Duncan k ratio t test (k ratio = 100) (SAS Institute, 1989).

At State College, the 11 accessions were evaluated for resistance to SWCB and FAW by using a single, 20-plant row for each insect. The accessions and a resistant (Mp704 × Mp707) and a susceptible (Ab24E × SC229) check were planted in the field on 23 April 1994. On 7 June, plants were infested with 30 neonates each, and the plants were visually rated on 21 June by using a scale of 0 (no damage) to 9 (extensive damage) (Davis and Williams, 1989; Davis et al., 1992).

At Columbia, the 11 accessions and a susceptible check, Ames 10583, were evaluated for resistance to WCRW. The accessions were grown at the Rollins

Table 1. Maize accessions from Peru resistant to first-generation European corn borer and evaluated for resistance to southwestern corn borer, fall armyworm, corn earworm, western corn rootworm, second generation European corn borer, and sugarcane borer

Entry	Southwestern corn borer[a]	Fall armyworm[a]	CEW 8 d larval wts (mg)[b]	CEW pupal wts (mg)[b]	CEW days to pupate[b]	Corn rootworm ($\bar{x} \pm$ SE)[c]	2nd gen. ECB[a] tunnel length (cm)[d]	Sugarcane borer[e] Ames	Sugarcane borer[e] Houma
PI 503720	7	7	225.3c	473.4a–d	13.6bc	2.67 ± 0.33	13.7b–e	6.5abc	6.3cd
PI 503722	7	6	180.2cde	450.3bcd	15.8abc	2.67 ± 0.33	23.4ab	6.0c	6.0cd
PI 503723	9	7	145.5def	435.4b–e	16.0abc	3.75 ± 0.28	33.7a	6.0c	6.5cd
PI 503725	8	7	101.2fg	473.8a–d	16.7abc	3.50 ± 0.28	15.3b–e	5.3cd	8.0ab
PI 503727	8	7	126.0efg	486.6abc	16.0abc	3.33 ± 0.33	6.2de	6.8abc	7.3bc
PI 503728	8	7	18.5h	359.3e	26.0a	3.25 ± 0.28	10.5cde	6.3bc	6.8bcd
PI 503731	8	7	146.9def	454.4bcd	15.6abc	3.67 ± 0.33	18.8bc	6.5abc	7.3bc
PI 503764	8	7	207.4cd	458.8a–d	16.6abc	3.75 ± 0.28	11.4cde	5.8cd	8.0ab
PI 503806	8	7	90.2fg	439.2b–e	16.8abc	3.25 ± 0.28	16.0bcd	5.5cd	6.3cd
PI 503849	8	8	59.8gh	—	—	4.50 ± 0.28	9.7cde	5.8cd	7.3bc
A-10623	9	7	102.1fg	393.0de	18.4abc	5.00 ± 0.28	5.1de	4.3d	5.5d
Mp704xMp707[f]	6	6	—	—	—	—	—	—	—
Ab24ExSC229[f]	8	8	—	—		—	—	—	—
B52[g]	—	—	—	—	—	—	3.4e	—	—
CI31A[h]	—	—	—	—	—	—	—	5.3cd	5.8d
WF9[h]	—	—	—	—	—	—	—	8.0a	9.0a
SEG[i]	—	—	329.4b	516.0ab	13.7bc	—	—	—	—
Z. Chico[i]	—	—	14.4h	352.7c	25.0ab	—	—	—	—
Bean diet[i]	—	—	595.0a	547.6a	12.2c	—	—	—	—
A-10583[j]	—	—	—	—	—	5.67 ± 0.28			

[a] Row rating in which 1 = no damage; 9 = extreme damage.
[b] Means followed by the same letter are *not significantly different according to the Waller-Duncan k-ratio* test ($P < 0.05$).
[c] Mean rating of 4 plants where 1 = no damage; 6 = three or more root nodes destroyed.
[d] Means followed by the same letter are not significantly different according to the LSD test ($P < 0.05$).
[e] 1 = no damage; 9 extreme damage.
[f] Checks used for SWCB and FAW tests.
[g] Check used for ECB test.
[h] Checks used for SCB tests.
Checks used for CEW tests.
[j] Check used for CRW test.

Table 2. Summary of resistance or intermediate resistance ratings of 11 European corn borer leaf-feeding resistant Peruvian maize accessions to several insect pests[a,b]

Accession no.	SWCB	FAW	CEW larval wt (mg)	CEW pupal wt (mg)	CW days to pupation	WCRW	ECB2	SCB	
								Ames	Houma
PI 503720				I	I	R	R	I	I
PI 503722		I		I	I	R		I	I
PI 503723				R	I	I		I	I
PI 503725			I	I	I	I	R	I	
PI 503727			I		I	I	R	I	
PI 503728			R	R	R	I	R	I	I
PI 503731				I	I	I		I	
PI 503764				I	I	I	R	I	
PI 503806			I	R	I	I		I	I
PI 503849			R			I	R	I	
A-10623			I	R	I		R	I	I

[a] R = resistant; I = intermediate resistance.
[b] SWCB = Southwestern corn borer; FAW = fall armyworm; CEW = corn earworm; WCRW = western corn rootworm; ECB2 = 2nd generation European corn borer; SCB = sugarcane borer.

Bottom site at the University of Missouri. Plots were infested with a tractor infester (Moellenbeck et al., 1994) when plants were at the 3- to 4-leaf stage at the rate of 600 eggs per 30.5 cm of row. Four plants from each accession were removed from the soil, washed, and rated using a root-damage rating scale of 1 to 6 (Hills and Peters, 1971) (1 = no damage or only slight feeding scars, 6 = 3 or more root nodes destroyed). Observations were made when infested larvae attained the late third-instar or early-pupal stage in a susceptible check plot. Data were analyzed with the ANOVA-1 program of MSTAT-C (MSTAT Development Team, 1989). Means from the 4-plant sample are reported as $\bar{x} \pm$ SE.

At Houma and at Ames, the 11 accessions were evaluated for leaf-feeding resistance by SCB. The accessions, plus an ECB-resistant check inbred CI31A and an ECB-susceptible check inbred WF-9 were planted in randomized block designs with four replications in rows 7.6 m long and 1.6 m apart. When the plants reached the V4-V6 stage of development, leaf whorls of 6 plants were infested with ca. 75 SCB neonate larvae by using the "bazooka" application technique developed by Mihm (1983). Damage was rated 3 weeks after infestation by using the ECB leaf-feeding rating scale developed by Guthrie et al. (1960). Data were analyzed with the ANOVA-2 program of MSTAT-C (MSTAT Development Team, 1989). When the F-value for treatments was significant ($P < 0.05$), means were separated with the least significant difference (LSD) test ($\alpha = 0.05$) included in the RANGE program of MSTAT-C.

Results and Discussion

Table 1 summarizes the results obtained from testing selected maize insect pests at each evaluation site. None of the 11 accessions was resistant to SWCB at Mississippi State; however, one accession, PI 503722, had intermediate resistance to FAW. The SWCB and FAW data are reported as "row" ratings since none of the individual plants within the rows showed any indication of resistance (resistance ratings would need to be from 1 to 3). At Tifton, two accessions, PI

503728 and PI 503849, produced 8-d CEW larval weights equal to the resistant check, and PI 503728 had larval development time and pupal weight equal to the resistant check. Three other PIs had pupal weights equal to the resistant check. There were two accessions, PI 503720 and PI 503722, resistant to WCRW root feeding at Columbia, whereas eight other accessions were considered intermediate in resistance to WCRW. At Ames, seven accessions had tunnel lengths caused by second-generation ECB feeding statistically equal to the B52 resistant check. All 11 accessions had intermediate resistance to SCB leaf feeding at Ames, whereas only 6 were intermediate in resistance at Houma.

Table 2 summarizes the overall resistant and intermediate resistance ratings from all the tests performed. There were 4 accessions resistant to 2 pests in addition to the ECB leaf feeding: PI 503720 was resistant to WCRW and ECB2, whereas PI 503728, PI 503849, and A-10623 were resistant to CEW and ECB2. PI 503720 also had intermediate resistance to CEW and SCB. Accessions PI 503728 and PI 503849 had intermediate resistance to WCRW and SCB. Accession A-10623 had intermediate resistance to CEW larval weight and days to pupation and to SCB. (Note: pupal weights and larval development times were not obtained for PI 503849 because of a shortage of silk).

Contemporary societies are concerned about the effects of pesticides on the environment and on the health of humans and other animals. Maize with resistance to several pests could be very useful from the perspective of both pest management and sustainable agriculture systems.

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